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Effects of YM90K, a selective AMPA receptor antagonist, on amygdala-kindling and long-term hippocampal potentiation in the rat

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Abstract

To investigate the role of α-amino-3-hydroxy-5-methyl-isoxazole-4-propionic acid (AMPA) type glutamate receptors in epileptic seizures, we examined the antiepileptogenic and anticonvulsant effects of YM90K [6-(1*H*-imidazol-1-yl)-7-nitro-2,3-(1*H*,4*H*)-quinoxalinedione hydrochloride], a potent and selective new AMPA receptor antagonist, in the rat amygdala-kindling model of epilepsy. Pretreatment with YM90K (7.5–30 mg/kg i.p.) markedly retarded the evolution of kindling. Once kindling was established, administration of YM90K (7.5–30 mg/kg i.p.) significantly and dose-dependently suppressed fully kindled seizures. The maximal effects were observed 15–30 min after injection. When the intensity of electrical stimulation was increased to twice the generalized seizure-triggering threshold, the anticonvulsant effects of YM90K were reversed, suggesting that they were due to elevation of the generalized seizure-triggering threshold. Furthermore, an anticonvulsant dose (15 mg/kg) of YM90K affected neither field potentials nor long-term potentiation in the hippocampus in vivo. These results indicate that AMPA receptors play an important role in the seizure expression mechanism and the development of kindling-induced epileptogenesis, and suggest the possible clinical usefulness of AMPA receptor antagonists as antiepileptic drugs.

Keywords: AMPA receptor antagonist; YM90K; Kindling; Long-term potentiation

1. Introduction

It is generally accepted that there are two major subtypes of inotropic glutamate receptors; the *N*-methyl-D-aspartate (NMDA) subtype and the non-NMDA subtype. Non-NMDA receptors are further classified into α-amino-3-hydroxy-5-methyl-isoxazole-4-propionic acid (AMPA) and kainate receptors (Meldrum, 1994; Löscher, 1998). A number of studies using animal models of epilepsy have indicated that NMDA and AMPA receptors play important roles in epileptogenesis and the expression of epileptic seizures (Meldrum, 1994; Löscher, 1998; Morimoto et al., 1998). Using the kindling model of epilepsy, we also have previously reported the roles of NMDA receptor antagonists, such as 3-(2-carboxypiperazine-4-yl)propyl-1-phosphonic acid (CPP), *cis*-4-phosphonomethyl-2-piperidine carboxylic acid (CGS19755), dizocilpine (MK801) and 7-chlorokynurenic acid. These studies showed that NMDA receptor antagonists significantly retard the development of kindling, but not the expression of fully kindled seizures (Sato et al., 1988; Morimoto et al., 1991; Namba et al., 1993). In contrast, 2,3-dihydroxy-6-nitro-7-sulfamoyl-benzo(*f*)-quinoxaline (NBQX), an AMPA receptor antagonist, not only retarded the development of kindling, but also suppressed fully kindled seizures (Namba et al., 1994). However, Dürmüller et al. (1994) could not confirm the effect of NBQX on the development of kindling. Therefore, in the present study, we investigated the antiepileptogenic and anticonvulsant effects of YM90K [6-(1*H*-imidazol-1-yl)-7-nitro-2,3-(1*H*,4*H*-quinoxalinedione hydrochloride], a potent and selective novel AMPA receptor antagonist, in the rat amygdala-kindling model of

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epilepsy. YM90K is as potent as NBQX in displacing [^{3}H]-AMPA binding to rat brain membrane and has little affinity for kainate and NMDA binding sites (Shimizu-Sasamata et al., 1996). In addition, to investigate its effects on synaptic transmission, the actions of YM90K on field potentials and long-term potentiation in the rat hippocampus were studied in vivo.

2. Materials and methods

2.1. Kindling

2.1.1. General procedure

Male Sprague–Dawley rats, weighing 260–350 g at the time of surgery, were used. They were housed under a 12-h/12-h light/dark cycle and allowed free access to food and water, except during the experimental sessions. The rats were anesthetized with sodium pentobarbital (50 mg/kg administered intraperitoneally, i.p.). A tripolar electrode was implanted stereotaxically into the left amygdala (2.5 mm posterior and 4.8 mm lateral to the bregma and 8.0 mm below the dura). The stereotaxic coordinates were determined using an incisor bar placed 3.3 mm below the interaural plane. The tripolar electrode consisted of three twisted Diamel-insulated Nichrome wires (0.18 mm in diameter). A screw electrode was placed into the right frontal skull to serve as a recording indifferent.

After a recovery period of 1–2 weeks, each rat was subjected to daily kindling stimulation sessions, each consisting of a 2-s train of 50-Hz biphasic square pulses at the intensity described below. The development of kindled seizures was assessed using a modification of the classification of Racine (1972). EEGs were recorded during all tests, between the remaining pole of the tripolar electrode and the skull screw electrode.

YM90K was dissolved in 1 N NaOH and diluted thereafter with 0.1% methylcellulose solution. The final pH of the YM90K solution was about 8.0–8.5.

2.1.2. Experiment 1: antiepileptogenic effects of YM90K on the development of amygdala-kindling

Twenty-five rats with tripolar electrodes implanted in their amygdala were used. After the recovery period, they were subjected to kindling stimulation. The afterdischarge threshold was determined in each rat by increasing the intensity of the stimulatory current by 25 μA at 20 min intervals until the animals produced afterdischarges (day 1). From days 2 to 30, the intensity of the kindling stimulating current was 150% of the afterdischarge threshold intensity. After the determination of afterdischarge threshold on day 1, the animals were divided into 4 groups, matched for seizure stages and afterdischarge durations. From days 2 to 9, the rats received YM90K 7.5 mg/kg ($N = 6$), 15 mg/kg ($N = 6$), 30 mg/kg ($N = 5$) or saline ($N = 8$) i.p. once per day, 30 min prior to each electrical kindling session. Drug administration was stopped on day 10, but daily kindling stimulation was continued until day 30.

The stages of kindled seizure development and increases in afterdischarge durations in the YM90K-treated groups were compared with those in the control (saline-treated) group.

2.1.3. Experiment 2: anticonvulsant effects of YM90K on previously amygdala-kindled seizures

Eight rats previously kindled via the left amygdala were used. Each rat received kindling stimulation at 150% of the afterdischarge threshold intensity once daily. Kindling stimulation was continued until the animals experienced at least five consecutive generalized convulsions (stage 5 seizures) over five successive days. The generalized seizure-triggering threshold was then determined in each rat by increasing the intensity of the stimulatory current by 25 μA at 20 min intervals until the animals produced generalized seizures. After stable stage 5 seizures had been induced by stimulation at the predetermined generalized seizure-triggering threshold intensity, drug experiments were performed as follows. Firstly, in order to determine whether the anticonvulsant effects of YM90K were dose-dependent, each rat was subjected to electrical stimulation at the individually determined generalized seizure-triggering threshold intensity 30 min after i.p. administration of YM90K (7.5, 15 or 30 mg/kg) or saline. Each drug or saline test was randomly sequenced and separated by at least 48 h. Secondly, in order to investigate the time course of the anticonvulsant effects of YM90K, the rats were subjected to electrical stimulation at the generalized seizure-triggering threshold 24 h before (predrug stimulation), 15 and 30 min after, and 1, 2 and 4 h after i.p. injection of YM90K (15 mg/kg). Finally, to determine the effect of YM90K on the generalized seizure-triggering threshold, the rats received electrical stimulation at twice the generalized seizure-triggering threshold, 30 min after i.p. administration of YM90K (15 mg/kg).

The anticonvulsant and antiepileptogenic effects of YM90K were assessed according to the kindled seizure stage scores and afterdischarge durations. Adverse effects, such as sedation, muscular hypotonia and ataxia, were also recorded, although the severity of YM90K-induced behavioral changes was not quantitatively scored in the present study.

2.2. Long-term potentiation

Ten male Sprague–Dawley rats (350–450 g) were anesthetized with urethane (1.4 g/kg, i.p.). The rectal temperature was monitored and was maintained at 36.5–37.0°C using a heating lamp.

In order to record evoked potentials in the hippocampus, a monopolar electrode (Teflon-coated stainless steel wire: 0.05 mm in diameter) was implanted into the dentate gyrus (3.2 mm posterior and 2.5 mm lateral to the bregma

and 2.5 mm below the dura). A stimulating monopolar electrode (Diamel-insulated Nichrome wire: 0.18 mm in diameter) was placed in the right entorhinal cortex (4.5 mm lateral to the lamda and approximately 2.7 mm below the dura) to stimulate the perforant path. The depth of the electrodes was adjusted to produce a maximum response. The stereotaxic coordinates were determined using an incisor bar, placed 3.3 mm below the interaural plane. A screw electrode was placed on the occipital skull. A small screw was attached to the frontal skull served as a ground.

Stimuli, included the tetanic stimulation, were applied between a monopolar electrode in the right entorhinal cortex and a screw electrode on the occipital skull. All stimuli were constant-current diphasic pulses, sequenced and timed by a programmable stimulator. Before the experiment, the current intensity required to evoke a population spike of >2 mV at a 250 μs pulse duration (150–500 μA) was determined. This intensity was used for the remainder of the experiment. For each response evoked in the dentate gyrus, the slope of the rising phase of the excitatory postsynaptic potential (EPSP) and the height of the population spike were measured.

Long-term potentiation was induced by 10 high-frequency (400 Hz) and high-intensity (250 μs pulses) train of impulses with a 20-ms duration, applied at 1-s intervals. To determine the synaptic strength and construct input/output (I/O) curves, the EPSPs were plotted against the log of the stimulatory intensity. I/O curve determination involved 32 single test pulses, ranging in intensity from 10 to 250 μs duration and delivered at 15-s intervals in an ascending–descending sequence. The first I/O curve was measured immediately before applying the tetanic stimulation, while the second was measured 45 min after application. Long-term potentiation was evaluated in terms of the percentage increase in the mean slope of the EPSP and the mean height of the population spike, calculated for five potentials evoked immediately before and five potentials evoked 45 min after the tetanic stimulation.

Test pulses of 250 μs duration were delivered at 30-s intervals before and after drug administration or the tetanic

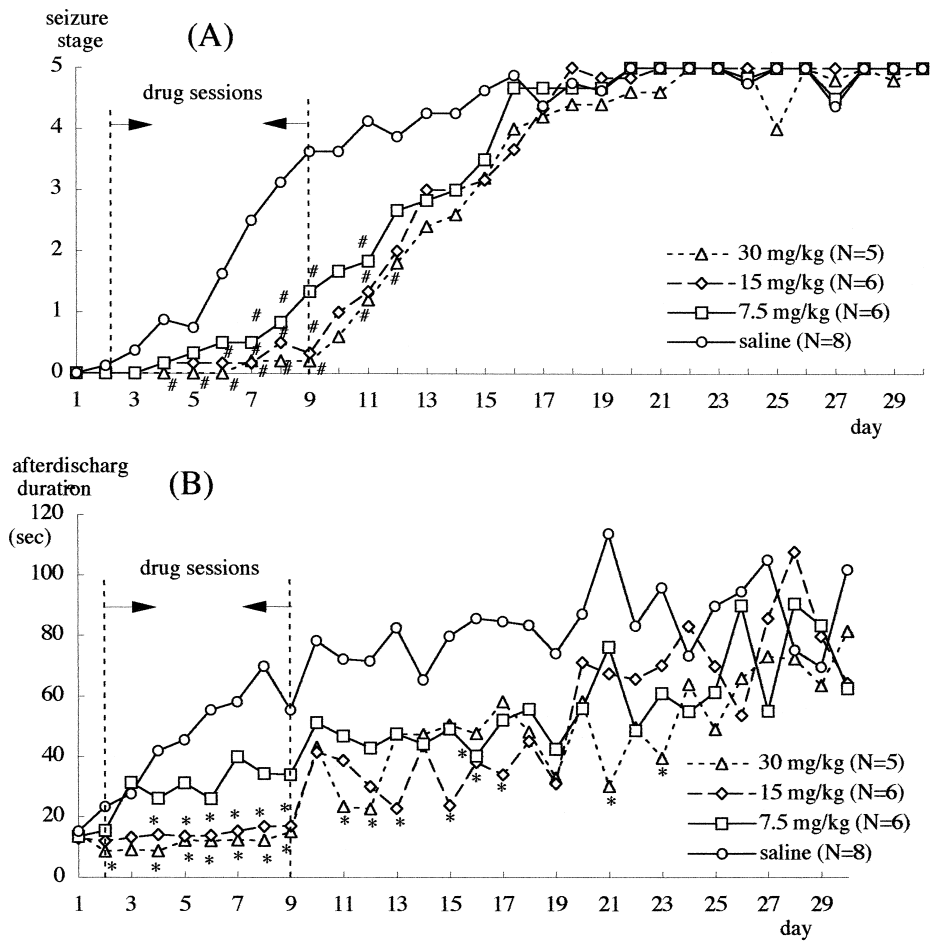

Fig. 1. Antiepileptogenic effects of YM90K on the development of amygdala-kindling. From day 2 to day 9, rats received YM90K (7.5, 15 or 30 mg/kg) or saline i.p., 30 min prior to each electrical kindling stimulation session. Pretreatment with YM90K significantly retarded the development of kindling (two-way ANOVA $P < 0.01$). (A): seizure stage scores; (B): afterdischarge durations; #$P < 0.05$ (Mann–Whitney U-test, as compared with control); *$P < 0.05$ (unpaired t-test, as compared with control).

Table 1
The mean number of stimulation sessions required to induce the first stage 2 and generalized stage 4 or 5 kindled seizure in the control (saline) and YM90K-treated groups

Treatment	No. of stimulation sessions required to reach each seizure stage	
	Stage 2 [range]	Stage 4 or 5 [range]
Control (saline) ($N = 8$)	7.0 [4–11]	9.8 [6–16]
YM90K		
7.5 mg/kg ($N = 6$)	10.8[a] [6–15]	14.5[a] [10–20]
15 mg/kg ($N = 6$)	13.0[b] [9–17]	14.5[a] [10–18]
30 mg/kg ($N = 5$)	12.8[b] [10–16]	15.6[a] [12–22]

[a] $P < 0.05$, [b] $P < 0.01$ (Mann–Whitney U-test, as compared with control).

stimulation. The tetanic stimulation was delivered 30 min after administering YM90K or saline and the evoked responses were recorded until 75 min after the injection. YM90K was used at the anticonvulsant dose, 15 mg/kg, determined by kindling experiments, and was prepared in the same way as described above. Both YM90K and saline were administered intraperitoneally.

2.3. Statistics

For all kindling experiments, afterdischarge durations are expressed as the mean ± S.E.M. and the seizure stage scores are expressed as mean (range). In Experiment 1, the seizure stages in the YM90K-treated groups were compared with the saline-treated (control) group using the Mann–Whitney U-test and the afterdischarge durations were compared using the unpaired t-test. A two-way repeated-measure ANOVA (analysis of variance) was used to evaluate repeated measurements of afterdischarge durations. In Experiment 2, the seizure stages were compared using the Wilcoxon test and the afterdischarge durations using the paired t-test. A one-way repeated-measure ANOVA was used to evaluate dose effects.

For the long-term potentiation experiments, all data are expressed as the mean ± S.E.M. A two-way repeated-measure ANOVA was used to evaluate repeated measurements of the evoked potentials.

Post-hoc analysis was performed using Fischer's protected least significant difference (PLSD). A P-value less than 0.05 was considered statistically significant.

3. Results

3.1. Kindling

3.1.1. Experiment 1

Fig. 1 shows the development of amygdala-kindled seizures in each group. Pretreatment with YM90K markedly retarded the evolution of kindling during the drug administration period. From day 2 to day 9, there were significant differences in afterdischarge duration between the four groups (two-way ANOVA, $P < 0.01$). After this time, kindling developed in the YM90K-treated rats and all rats displayed stage 5 seizures by day 30.

Table 1 shows the mean number of stimulation sessions required to induce the first stage 2 and generalized stage 4 or 5 kindled seizures in the control and YM90K-treated groups. The number of stimulation sessions necessary to induce these stages was significantly greater for each YM90K-treated group than for the control group. When stimulation during the drug administration period was excluded, the numbers of sessions required to induce the first generalized stage 4 or 5 seizures were 9.8 (control group),

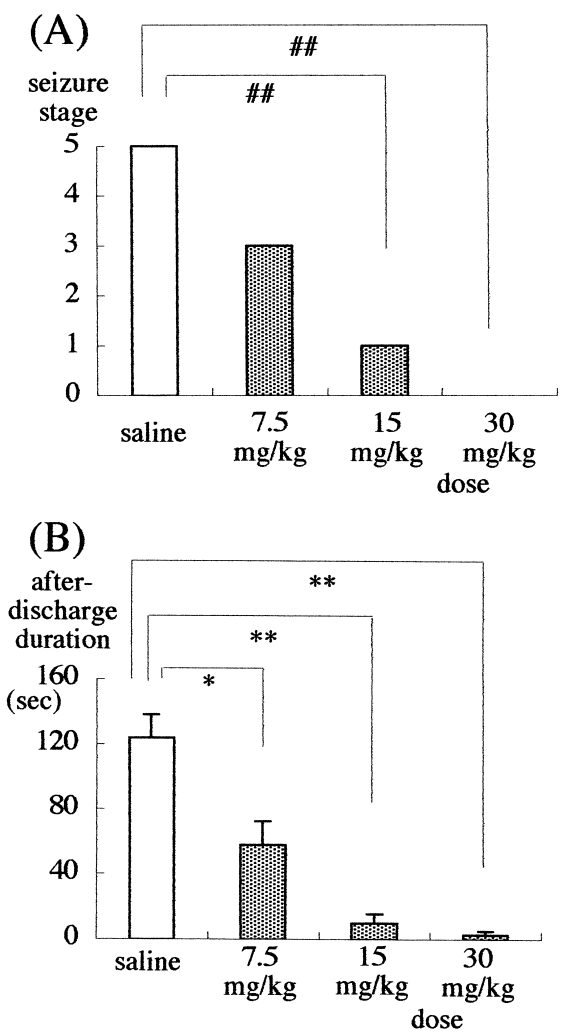

Fig. 2. The dose-dependency of the anticonvulsant effects of YM90K on fully kindled amygdala seizures. Rats ($N = 8$) were subjected to electrical stimulation at the same intensity as the generalized seizure-triggering threshold, 30 min after i.p. administration of YM90K (7.5, 15 or 30 mg/kg) or saline. Pretreatment with YM90K significantly reduced both the seizure stages attained and the afterdischarge durations in a dose-dependent manner (one-way ANOVA $P < 0.01$). (A): seizure stage scores; (B): afterdischarge durations; $\#\#P < 0.01$ (Wilcoxon test, as compared with control); $^{*}P < 0.05$, $^{**}P < 0.01$ (paired t-test, as compared with control).

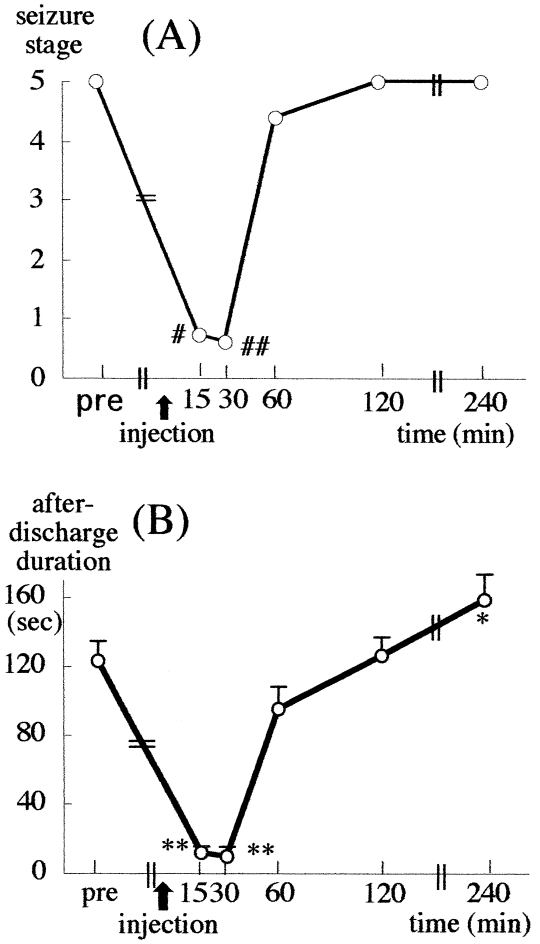

Fig. 3. The time course of the anticonvulsant effects of YM90K on fully kindled amygdala seizures. Rats ($N = 8$) were subjected to electrical stimulation at the same intensity as the generalized seizure-triggering threshold 24 h before (predrug stimulation), 15 and 30 min after, and 1, 2 and 4 h after i.p. injection of YM90K (15 mg/kg). Both the seizure stages attained and the afterdischarge durations were significantly reduced 15 and 30 min after the injection. (A): seizure stage scores; (B): afterdischarge durations; $\#P < 0.05$, $\#\#P < 0.01$ (Wilcoxon test, as compared with preinjection); $^{*}P < 0.05$, $^{**}P < 0.01$ (paired *t*-test, as compared with preinjection).

6.5 (7.5 mg/kg YM90K group), 6.5 (15 mg/kg YM90K group) and 7.6 (30 mg/kg YM90K group), respectively. The results for each YM90K-treated group were not significantly different from those for the control group (Mann–Whitney *U*-test), suggesting that YM90K inhibited kindling-induced elongation of the afterdischarge duration during the drug administration period. The waveform, such as the frequency and amplitude, of the afterdischarges in the YM90K-treated group almost did not change during the drug administration period, whereas those in the control group greatly increased during the period.

3.1.2. Experiment 2

Fig. 2 shows the dose-dependency of the anticonvulsant effects of YM90K on previously fully kindled amygdala seizures. Pretreatment with YM90K markedly and significantly reduced both the seizure stages attained and the afterdischarge durations of fully kindled seizures in a dose-dependent manner (one-way ANOVA, $P < 0.01$). None of the rats treated with YM90K (15 or 30 mg/kg) showed stage 5 seizures, and their afterdischarge durations were reduced or afterdischarges were incompletely evoked. Even when full afterdischarges were evoked, their frequency and amplitude decreased.

The time course of the anticonvulsant effects of YM90K (15 mg/kg) on fully kindled seizures is shown in Fig. 3. The maximal effects on both seizure stages and afterdischarge durations were observed 15–30 min after the injection. At these, both the seizure stages and the afterdischarge durations were significantly reduced compared with the baseline values. After 1 h, the anticonvulsant effects disappeared.

Table 2 shows the effects of YM90K on the generalized seizure-triggering threshold for fully kindled seizures. At the generalized seizure-triggering threshold intensity, YM90K (15 mg/kg) completely abolished the induction of afterdischarges in five out of eight rats. Only one rat showed generalized seizures. However, the effects of YM90K were reversible, as generalized seizures returned in six out of eight rats when the intensity of the current was raised to twice the generalized seizure-triggering threshold. The two remaining rats showed partial seizures with short afterdischarge durations.

Table 2
Effects of YM90K on the GST for fully kindled amygdala seizures. Rats received electrical stimulation at twice the GST, 30 min after i.p. administration of YM90K (15 mg/kg)

Treatment/stimulus intensity	Seizure stage [range]	AD duration (mean ± S.E.M.)	No. of rats showing	
			Generalized seizures	No ADs
Control (saline)				
GST ($N = 8$)	5.0 [5–5]	123.8 ± 14.2	8/8	0/8
YM90K (15 mg/kg)				
GST ($N = 8$)	0.6[a] [0–4]	9.8 ± 5.5[c]	1/8	5/8
Twice the GST ($N = 8$)	4.1 [1–5]	78.9 ± 13.3[b]	6/8	0/8

AD: afterdischarge; GST: generalized seizure-triggering threshold.
[a] $P < 0.01$ (Wilcoxon test, as compared with control).
[b] $P < 0.05$, [c] $P < 0.01$ (paired *t*-test, as compared with control).

Intraperitoneal injection of 15–30 mg/kg YM90K induced various behavioral changes, including sedation, muscular hypotonia and ataxia, which reached their peak intensity 10–30 min after injection. However, administration of 7.5 mg/kg YM90K produced very few behavioral changes.

3.2. Long-term potentiation

As shown in Fig. 4, the administration of 15 mg/kg YM90K had no significant effect on field potentials in the hippocampus in vivo. There were no general changes in either the EPSP or the population spike, before and after injection (two-way ANOVA).

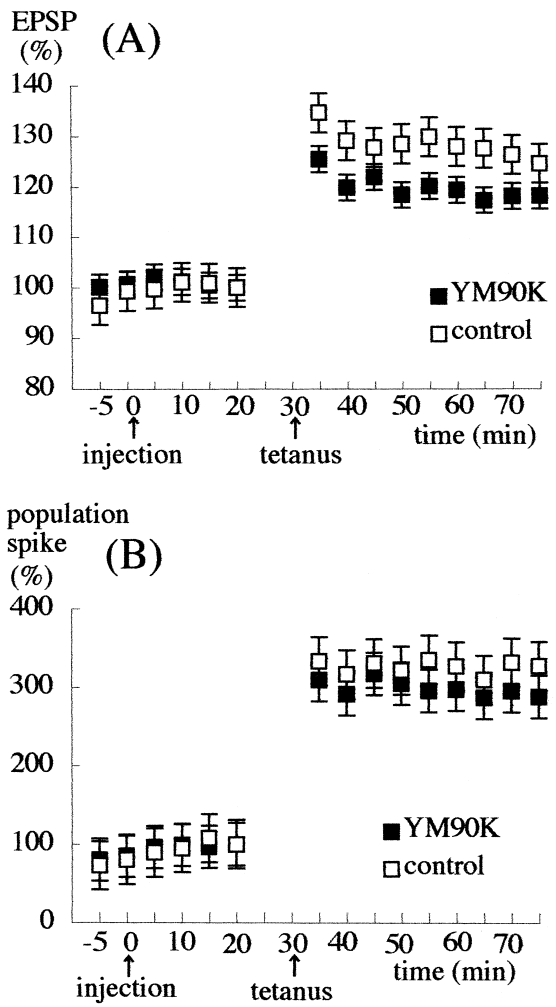

Fig. 4. The effects of YM90K on field potentials and long-term potentiation in the hippocampus in vivo. The tetanic stimulation was delivered 30 min after the administration of YM90K (15 mg/kg) or saline, and the responses evoked were recorded until 45 min after the onset of tetanus. YM90K had no significant effect on field potentials in the hippocampus. In contrast, the application of tetanic stimulation produced an increase in evoked potentials in YM90K-treated as well as in control rats. However, there were no significant differences between the YM90K-treated and control groups in terms of the EPSP and the population spike (two-way ANOVA). (A) EPSP; (B) population spike.

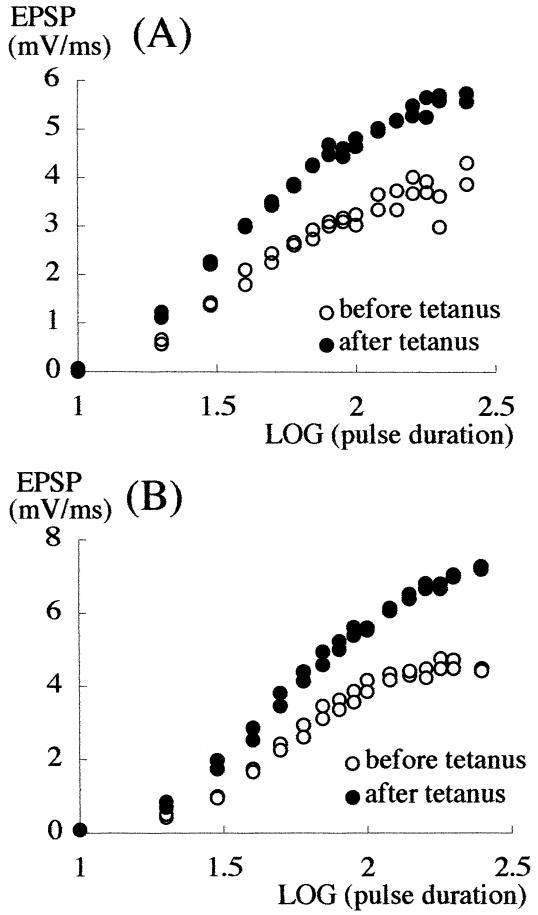

Fig. 5. Examples of I/O curves generated from a stimulus intensity series before and after tetanic stimulation in control (A) and YM90K-treated rats (B). The rats in both the YM90K-treated and control groups showed a substantial increase in the slope of the second I/O curve (recorded 45 min after the onset of tetanus) when compared with the first I/O curve (recorded immediately before the onset of tetanus).

The application of tetanic stimulation produced an increase in evoked potentials in the YM90K-treated as well as the control rats. In both groups, the EPSP and the population spike, 5–45 min after the onset of tetanus, were significantly potentiated compared with those recorded before the onset of tetanus. Forty-five minutes after the onset of tetanus, the mean ± S.E.M. potentiation of the EPSP and the population spike was 24.6 ± 5.1% and 227.9 ± 94.4% in the control group, 18.2 ± 4.4% and 188.7 ± 36.3% in the YM90K-treated group. However, there were no significant differences in terms of the EPSP and the population spike between the YM90K-treated and control groups (two-way ANOVA).

As shown in Fig. 5, all rats in the YM90K-treated group, as well as those in the control group, showed a substantial increase in the slope of the second I/O curve (recorded 45 min after the onset of tetanus) when compared with the first I/O curve (recorded immediately before the onset of tetanus).

To summarize the results, the administration of YM90K had no significant effects on field potentials or long-term potentiation in the hippocampus in vivo.

4. Discussion

The results of experiment 1 clearly demonstrate that YM90K possesses potent antiepileptogenic activity. In the development of kindling, Repetitive activation of NMDA receptors has been generally accepted to be important in the development of kindling (Sato et al., 1988; Morimoto et al., 1991; Namba et al., 1993; Meldrum, 1994; Löscher, 1998). However, with regard to the role of AMPA receptors, conflicting results have been obtained in studies using NBQX (Dürmüller et al., 1994; Namba et al., 1994). The results of the present study are almost identical to those of our previous study of NBQX (Namba et al., 1994), suggesting that AMPA receptor do indeed participate in the development of kindling. Ernfors et al. (1991) have also indicated that AMPA receptors play an important role in the development of kindling, based on in situ hybridization of neurotrophic factors. Investigators using other models of epilepsy have also reported that AMPA receptors are probably involved in the initiation of seizure activity (Young and Dragunow, 1994).

In experiment 1, the frequency and amplitude of the afterdischarges in the YM90K-treated group were not increased as much as in the control group. After drug administration ceased, the number of stimulation sessions required to complete kindling remained the same as in controls, suggesting that the antiepileptogenic action of YM90K results from inhibition of seizure activity within the stimulated region, and in consequence, development of the epileptic neural circuit was suppressed.

Experiment 2 clearly showed that YM90K has a potent anticonvulsant effect on seizures previously kindled via the amygdala in rats. This is consistent with the effects of other AMPA receptor antagonists, such as NBQX, which have been studied in various experimental models of epileptic seizures in vivo (Yamaguchi et al., 1993; Meldrum, 1994; Namba et al., 1994; Swedberg et al., 1995; Löscher, 1998). Furthermore, YM90K has also been observed to have anticonvulsant effects in other experimental epilepsy models, such as audiogenic seizures in DBA/2 mice and the partially kindled hippocampal seizure model (Shimizu-Sasamata et al., 1996; Katsumori et al., 1998). All these data support the theory that AMPA receptors play a critical role in the seizure expression mechanism, not only in kindled seizures, but also in various other experimental epilepsy models.

The anticonvulsant effects of YM90K also appear to result from inhibition of seizure activity in the stimulated region (elevation of seizure threshold). An increase in the intensity of stimulation reduced or even reversed the effects of YM90K, especially in terms of the seizure stages attained. In this respect, the effects of YM90K are similar to those of other antiepileptic drugs, such as phenytoin, carbamazepine and lamotrigine (Morimoto et al., 1997; Otsuki et al., 1998). It has been suggested that the principal mechanism underlying the anticonvulsant activity of these compounds is elevation of the seizure threshold in a kindled epileptogenic focus. However, the effects of YM90K seen in the present study are different from those which we previously reported for NBQX (Namba et al., 1994). In the NBQX study, elevation of the intensity of stimulation did not reverse the effects on amygdala-kindled seizures and produced few changes in the waveforms of the afterdischarges. We therefore suggested that the anticonvulsant effects of NBQX are due to blockade of the propagation pathway of afterdischarges, especially within and/or around the amygdala focus.

The reasons for this discrepancy in the mechanisms of action of YM90K and NBQX are unclear. The protocol used during the NBQX experiment was almost the same as that of the present study. However, the electrical stimulation used in the NBQX study consisted of sine waves, while square waves were used in this study. This subtle difference in the form of stimulation may have an influence on the results. There is, however, a recent review which indicates that the anticonvulsant effects of NBQX result from suppression of both the initiation and propagation of seizure activity (Löscher, 1998). In contrast, the anticonvulsant effects of YM90K appear to result mainly from inhibition of the seizure initiation process. The propagation of seizure activity might, however, have also been suppressed by YM90K, since some animals showed partial seizures at an increased stimulatory intensity in this study.

It is quite clear that activation of AMPA receptors is important for seizure expression. Indeed, AMPA receptors are probably more important than NMDA receptors for the expression of kindled seizures, because NMDA receptor antagonists, such as MK801, show relatively weak anticonvulsant activity in the kindling model (Sato et al., 1988; Morimoto et al., 1991; Meldrum, 1994; Löscher, 1998). Moreover, in a study of epileptic seizures evoked by application of a GABA receptor antagonist to the 'area tempestas', AMPA receptors were reported to have a more important role than NMDA receptors in limbic seizure expression (Tortorella et al., 1997).

In the long-term potentiation experiments, it was evident that an anticonvulsant dose of YM90K did not change the hippocampal field potentials in vivo when the rats were anesthetized with urethane. Furthermore, YM90K did not affect long-term potentiation in the hippocampus. The dose we used for the long-term potentiation experiments was 15 mg/kg, at which dose both kindling development and kindled seizures were significantly suppressed. Therefore, it is unlikely that the antiepileptogenic and anticonvulsant effects of YM90K are due to a reduction in synaptic transmission to the sites relevant for long-term potentiation induction.

These results on long-term potentiation are also quite consistent with those of our previous experiment using NBQX under almost the same conditions (Sato et al., 1995). We have also shown that NMDA receptor antagonists, such as MK801, CPP and CGS19755, almost completely blocked long-term potentiation in vivo (Morimoto et al., 1991). Furthermore, it has been reported that YM90K dose not impair spatial cognition in rats during a radial maze task (Li et al., 1996, 1997). Since long-term potentiation is considered to be a model of memory or a similar process (Bliss and Collingridge, 1993), we suggest that YM90K may be safer than NMDA receptor antagonists, at least in respect of its influence on the brain functions involved in memory. However, further studies will be necessary to determine whether YM90K is still harmless to memory functions after repeated administration.

YM90K has also been reported to produce no neurotoxic effects on cerebrocortical neurons in rats (Izumisawa et al., 1995). Although the dose required to show the antiepileptogenic and anticonvulsant effects in humans is unclear, it was reported that YM90K induced neither significant adverse reactions nor severe abnormalities during physical and laboratory examinations in healthy humans (Umemura et al., 1997a). In addition, some reports have indicated that acute application of YM90K has neuroprotective effects in focal and global ischemia models (Shimizu-Sasamata et al., 1996; Yatsugi et al., 1996; Kawasaki-Yatsugi et al., 1997; Umemura et al., 1997b; Yao et al., 1997). However, high or moderate doses of YM90K induced behavioral changes, including sedation, muscular hypotonia and ataxia, during our present experiments. In addition, the effects of YM90K were short-lived. In this context, YM90K may be useful for the treatment of acute epileptic conditions such as status epilepticus. Moreover, it might prevent pathological neuronal plasticity resulting from status epilepticus, since it showed an antiepileptogenic effect in this study and protected hippocampal neurons during kainate-induced generalized seizures (Shiraishi, 1997). YM90K could be clinically useful, since even a low dose exhibited significant, although weak, effects against kindling without detectable behavioral adverse effects.

In conclusion, YM90K exhibits potent antiepileptogenic and anticonvulsant effects in the kindling model of epilepsy. This suggests that AMPA receptors play a crucial role in the mechanism underlying seizure generation and are also involved in kindling-induced epileptogenesis. Furthermore, it is likely that AMPA receptors are more important than NMDA receptors in seizure expression. These findings indicate the possible clinical usefulness of AMPA receptor antagonists as antiepileptic drugs.

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